

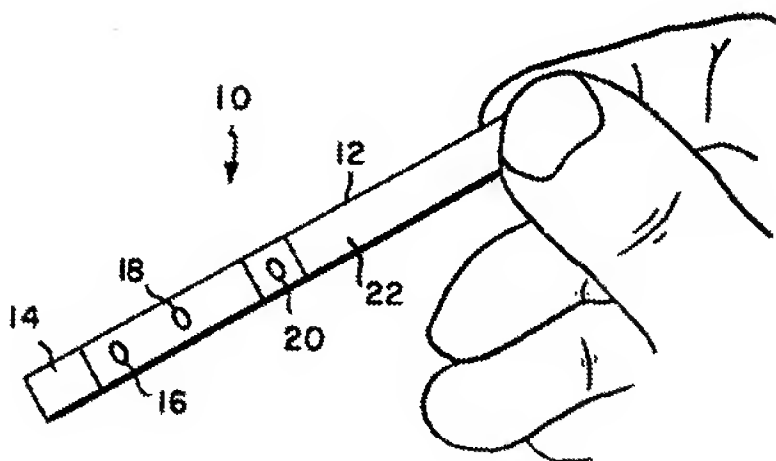


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(54) Title: ASSAYS FOR COMPOUNDS IN CONSUMABLE ITEMS**(57) Abstract**

The present invention relates to simple indicators that allow a consumer to determine the presence or absence of particular compounds, such as caffeine and lactose, in foods and beverages. The indicators include portions which provide rapid visual indication of the presence (or absence) of the subject compounds. The indicators are sized to permit them to be carried within a pocket or purse allowing portability and discreet use.



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ASSAYS FOR COMPOUNDS IN CONSUMABLE ITEMS

Field of the Invention

This invention is in the field of assays for specific compounds in consumable items such as foods and beverages. In particular, the invention relates to simple assays by which an individual can determine the presence of caffeine or lactose in consumable items using a simple non-automated technique.

Background of the Invention

The detection of compounds in various foods and beverages is a well-developed technology practiced in analytical chemistry laboratories. Producers of foods and beverages typically require laboratory services to determine the quality and safety of their products, and also to insure that their products remain substantially uniform regardless of raw materials, seasonal variations, etc.

It is relatively easy to determine the presence of a wide variety of compounds using analytical chemistry techniques. However, such methods often are not available, or practical, for individual consumers seeking to determine the presence or absence of certain compounds in their food and beverages. For example, although "decaffeinated" coffees, teas, and soft drinks have become increasingly popular over the past several years, the average consumer has no way of verifying the absence (or presence) of caffeine in such beverages when receiving them in restaurants and other public and private settings. It should be understood that the term "decaffeinated" as used herein is a relative term referring to beverages having a reduced caffeine content. For example, a cup of coffee typically contains about 100 mg of caffeine (approximately 400mg/L) whereas a cup of decaffeinated coffee typically contains about 4 mg of caffeine (approximately 16mg/L), and a cup of tea typically contains about 35 mg of caffeine (approximately 140mg/L) whereas a cup of decaffeinated tea typically contains less than about 2 mg of caffeine (approximately 8mg/L). Additionally, since most people find caffeinated and decaffeinated beverages to be very similar in taste, a consumer has very limited ability to determine whether a beverage received in a restaurant or other setting lacks (or contains) caffeine. Thus, if a group of diners orders a combination of caffeinated and decaffeinated beverages, it becomes very difficult to assure that the server properly distributed the caffeinated and decaffeinated beverages to the proper persons.

In a related example, a significant percentage of the population lacks the enzymes necessary to digest lactose. This "lactose-intolerance" increases dramatically with age, starting

in infancy and progressing through adolescence. The prevalence of lactose-intolerance is known to vary according to ethnicity. For example, virtually all Vietnamese adults are lactose-intolerant, as are approximately 95% of Native American adults, 65% of African-American adults, 22% of Caucasian adults, and 7% of Northern European adults.

5 Lactose-intolerant individuals often experience a significant level of gastrointestinal distress if they consume milk or other products containing lactose. Although milk and other food and beverage products that are said to have reduced lactose content or to be free of lactose are available in supermarkets, grocery stores, restaurants and the like, those consuming such products cannot guarantee that the products consumed actually are free of lactose until a

10 significant time period has elapsed following consumption of the products.

Additionally, some foods (such as sorbet) may or may not contain milk or milk products, and it is often very difficult to tell if such are present at all, let alone in a lactose-reduced form.

Numerous methods are known to analytical chemists for determining the presence of caffeine and lactose in various media. For example, in the case of caffeine, the scientific

15 literature includes methods such as electrometric determination in which a caffeine-specific electrode is prepared from a caffeine-picrylsulfonate ion-pair complex dissolved in octanol; fluorimetric determination in which a buffered solution of caffeine is oxidized with N-bromosuccinimide and then reacted with dimethyl o-phenylenediamine followed by a fluorescence measurement at 480 nm; colorimetric determination in which an ethenolic solution

20 of caffeine is oxidized by potassium bromate, dried and then redissolved in dimethylformamide followed by an absorbance measurement at 500 nm; Fourier Transform Infrared Spectrophotometry (FTIR); thin-layer/gas chromatography; enzyme-linked immunosorbent caffeine assays in which a caffeine-containing sample of plasma or serum is dissolved in a buffered solution and incubated in a vessel where it competes with peroxidase-labeled caffeine

25 for the binding sites on caffeine antibodies followed by detection of a visible color change with the addition of o-phenylenediamine; immunoassay of theophylline with cross-sensitivity for caffeine; and immunoliposome assay of theophylline with cross-sensitivity for caffeine.

Likewise, numerous assays exist for determining the presence of lactose in various media. These include the lactose enzyme electrode in which a lactose specific electrode is

30 prepared using either galactose oxidase or a beta galactosidase/glucose oxidase combination in conjunction with an H_2O_2 electrode, whereby lactose reacts with the galactose oxidase or galactosidase/glucose oxidase combination to liberate peroxide which produces an

electrochemical response; lactose determination by micro-calorimetry in which the reaction of lactose with beta galactosidase (lactase) to produce glucose and galactose is used; Benedict's reagent in which a cupric citrate alkaline solution is used to detect reducing sugars; and the method specified in U.S. Patent No. 3,814,668 for the semi-quantitative determination of glucose
5 in a sample fluid in which a sample containing glucose reacts with glucose oxidase to liberate oxygen, the peroxidase accelerates the release of oxygen (peroxide), and the peroxide reacts with potassium iodide releasing iodine and producing a color change to brown.

The problem inherent in most test procedures for lactose is that the majority are not lactose-specific. Rather, most will also react with glucose. Furthermore, each of the caffeine
10 and lactose assays described above requires the use of laboratory instrumentation and techniques in carrying out the assay and, as such, they are not well suited for consumer use.

In view of the above, a need exists for a simple, inexpensive test that can be used by a consumer to determine the presence (or absence) of compounds such as caffeine and lactose in consumable food and beverage products.

15 Furthermore, a need exists for a simple assay which can be carried out without the need for instrumentation or laboratory techniques.

A need also exists for an assay which can determine the presence (or absence) of compounds such as caffeine and lactose in a food or beverage portion without adulterating the portion (or at least a significant amount thereof).

20 A need also exists for a simple assay which is easily portable and may be performed quickly and discreetly.

Summary of the Invention

The present invention relates to simple assays to detect the presence (or absence) of materials such as caffeine and lactose in consumable items such as foods and beverages. The
25 assay materials may be easily carried in a pocket or purse, and the assay can be performed in a simple and discreet manner. Each assay comprises an indicator, in the form of a wick, a capillary tube, a dipstick or the like having appropriate reagents thereon, which can be contacted with a sample taken from a food or beverage portion and can provide rapid visual feedback indicating the presence (or absence) of the subject compound, or in some cases, its concentration.

30 In one preferred embodiment, caffeine and lactose indicators are enclosed in a matchbook-type package which includes a plurality of caffeine indicators, lactose indicators, or a combination thereof. Such a construction offers a simple, disposable, inexpensive package that

is easily used by a consumer. In that embodiment, a consumer ordering a cup of, for example, decaffeinated cappuccino or caffe latte, would open the matchbook, tear off indicators for caffeine and lactose, and briefly contact those indicators with a sample of the beverage. Shortly thereafter, colored portions on the indicators could be viewed to determine the presence or
5 absence of caffeine and lactose in the beverage.

Brief Description of the Drawings

FIG. 1 is a schematic depiction of one embodiment of an indicator for caffeine;

FIG. 2 is a schematic depiction of one embodiment of an indicator for lactose;

FIG. 3 is a schematic depiction of a package containing a plurality of indicator strips.

10 FIG. 4a and 4b depict a housing containing an indicator and illustrate its method of use.

Detailed Description of the Invention

The present invention provides a simple assay for determining the presence (or absence) of compounds such as caffeine or lactose in a food or beverage sample. In one embodiment, the subject assays provide a system using a sample "dipstick" which allows a consumer to verify
15 whether the target species are present simply by contacting the dipstick with the food or beverage sample and observing an indication, typically a colorimetric indication, thereon.

In the case of caffeine, determination is made using an immunoassay. In particular, caffeine-specific antibodies, raised in various laboratory animals, can bond to a specific number of caffeine molecules at a limited number of binding sites. By simultaneously introducing a
20 caffeine sample taken from a beverage and a control standard such as a labeled caffeine sample to a caffeine-specific antibody, a competitive assay results in which the labeled and unlabeled caffeine molecules compete for the available binding sites on the antibody. Any unbound, labeled caffeine is then available to contact an indicator to provide an indication of the presence of caffeine in the consumable sample. For example, if a control standard of caffeine is labeled
25 with an agent that causes a color change in an indicator and introduced to a caffeine-specific antibody simultaneously with a beverage sample which does not contain caffeine, all available binding sites on the antibody would be occupied by the labeled caffeine, thereby leaving few of the labeled moieties available in an unbound form to induce a color change in the indicator. Thus, little color change in the indicator would be expected to develop. Likewise, if equal
30 concentrations of labeled and unlabeled caffeine were provided to the antibodies, about half of the maximum possible color change would be expected, since about half of the antibody binding sites would be occupied by labeled caffeine with the other half being occupied by unlabeled

caffeine. The unbound labeled caffeine complexes would remain available to induce a visual change in the indicator. Thus, as the concentration of caffeine in the sample increases, the number of binding sites on the antibody occupied by labeled-caffeine decreases, thereby freeing more labeled caffeine for interaction with the indicator. The result is a greater color change on the indicator. In one preferred embodiment, these effects can be produced by labeling caffeine with peroxidase and using o-phenylenediamine or other suitable indicating reagents to detect the presence of unbound, labeled caffeine.

One preferred embodiment of a caffeine indicator is depicted in FIG. 1. In that Figure, the indicator 10 comprises a test strip 12 formed of a material having the ability to wick fluids. One preferred material is filter paper. The indicator 10 includes five separate portions: a sample liquid portion 14, a labeled caffeine portion 16, a caffeine antibody portion 18, an indicator portion 20, and a grip portion 22.

In use, the liquid sample portion 14 is contacted with a liquid, such as coffee, to be tested. The portion 14 may either be immersed in the coffee or the coffee may be dripped onto the liquid sample portion using a spoon, dropper or the like. It is preferred that only the liquid sample portion be contacted with the beverage being tested. If the entire indicator is immersed in the beverage, caffeine present in the sample will be able to bind with the antibody faster than the labeled caffeine, thereby providing inaccurate results. Additionally, such immersion raises the possibility of introducing unwanted chemical reagents into the beverage. Each of these effects could be avoided by providing a relatively impermeable sheath or jacket around the indicator except in the sample liquid portion 14. Suitable jacket materials include any of a wide variety of transparent polymeric materials approved by regulatory agencies for food and beverage contacting applications.

As the sample liquid wicks upward along the indicator strip, it first encounters and mixes with a labeled caffeine, such as peroxidase-labeled caffeine, that has been impregnated into the wickable material in the labeled caffeine portion 16. The labeled caffeine mixes with any caffeine contained in the liquid sample and is carried upward along the wick as the wicking process continues. As the sample liquid enters the caffeine antibody portion 18, labeled and non-labeled caffeine compete for binding sites on the caffeine antibodies. As noted above, if the sample liquid is substantially free of caffeine, substantially all of the labeled caffeine will become bound to the caffeine antibodies. In contrast, as the amount of caffeine in the sample liquid increases, a greater amount of the labeled caffeine will be unable to attain a binding site on

the antibody and will be free to continue wicking along the indicator strip.

The sample liquid, now containing a lower percentage of labeled caffeine as a result of interaction with the caffeine antibodies continues to wick along the indicator strip into the indicator portion 20. Based upon the amount of labeled caffeine interacting with the indicator (o-phenylenediamine or the like), the color of the indicator will react. If the sample is relatively free of caffeine, only a small amount of, for example, peroxidase-labeled caffeine will be available to interact with the indicator and thus, no significant color change will occur in the indicator. In contrast, if the sample beverage contains a significant amount of caffeine, a greater amount of unbound peroxidase-labeled caffeine will be available for interaction with the indicator and the color change of the indicator will be significant. By examining the color response of the indicator, the consumer thus has an indication as to whether the sample beverage contains or lacks caffeine. Likewise, by examining the amount of color change, the consumer can determine not only the presence (or absence) of caffeine, but the relative amount present as well. The entire indicator strip can be easily manipulated by the consumer via the gripping portion 20.

It should be noted that the present invention is not intended to be limited to the specific competitive assay method described above. For example, other approaches may be employed including apoenzyme reactivated immunoassay (ARIS) and "enzyme channelling". In the ARIS assay, caffeine is linked to the enzyme prosthetic group flavin adenine dinucleotide (FAD), the co-factor for glucose oxidase. The FAD-caffeine complex competes with free caffeine for binding to anti-caffeine antibodies. As with the competitive assay described above, the higher the content of caffeine in the beverage, the more unbound FAD-caffeine is available for interaction with apoglucose oxidase, thereby converting it to active enzyme to metabolize glucose in the presence of peroxidase and tetramethylbenzidine as color reagents. The result is an increasing blue color with increasing caffeine concentration. In this embodiment, the test device may comprise a plastic strip having filter paper impregnated with an aqueous phase followed by drying and secondary application of an organic phase. The aqueous dip includes anti-caffeine antibodies, apoglucose oxidase, anti-glucose oxidase (for enzyme stabilization), peroxidase and glucose. The organic phase contains FAD-caffeine and tetramethylbenzidine or similar color reagents. Further description of the ARIS procedure may be found in Greenquist, AACC TDM-I, Vol. 6, No. 6, pp. 1-8, December, 1984, the teachings of which are incorporated herein by reference. Of course other enzyme systems may be employed.

In connection with ARIS arrays, a multilayer immunochemical dipstick employing multifilm technology may be used. The result is that the reagents are spatially separated in various parts of the reaction. Such a device is depicted schematically on page 7 of the Greenquist reference.

5 In the enzyme channelling reaction, caffeine linked to peroxidase is again used. Free caffeine in a beverage competes with caffeine-peroxidase for binding to caffeine antibodies which are linearly spaced along a wicking path. The higher the amount of caffeine in the sample, the greater the competition for binding sites along the wicking path. As a result, if the sample has a high caffeine concentration, labeled caffeine is likely to wick further along the path. By
10 determining the length of travel of the labeled caffeine, the relative caffeine concentration of the sample can be determined. The assay can be achieved in a multilayer dipstick model, sequential impregnated papers on a plastic dipstick, or within a thin plastic "thermometer"-type device where the height of the color (i.e., distance traveled by free caffeine-peroxidase) is proportionate to the amount of caffeine in the beverage. Further description of the enzyme channeling assay
15 may be found in Wagman et al., AACC TDM-T, Vol. 7, No. 8, pp. 1-6, February, 1986, the teachings of which are incorporated herein by reference.

For each assay, multiple impregnated paper strips aligned along a plastic support above a chemical-free "wick" may be employed. Multifilm layer technology, or a defined volume capillary tube capable of being immersed in the beverage may be used as well. In the latter case,
20 the capillary tube can have the appropriate reagents immobilized upon the interior walls of the lumen. The result is a hollow "thermometer"-type device. The test strips or "thermometers" may be packaged either in a matchbook like device or in a small container readily carried by a consumer. Depending upon size and configuration of each device, a small premeasured plastic sampling spoon may also be provided or physically incorporated into the assay device. Also, a
25 strip of a support material such as a semi-rigid plastic may be affixed to each indicator to provide some level of physical support and ease of manipulation.

In the case of the lactose-indicating devices, the preferred embodiment includes indicators for the presence of both glucose and lactose. This is because many of the lactose indicators are also sensitive to the presence of glucose, but not necessarily vice-versa. Thus, a
30 glucose indicator can be compared (as a control) to a lactose indicator with a difference between the two being indicative of the presence of lactose. In one preferred embodiment shown in FIG. 2, the lactose indicator 50 comprises a dipstick 52 having a glucose test portion 54 and a

glucose/lactose test portion 56 mounted at the end of a plastic strip. Each test portion can consist of an absorbent paper impregnated with an indicator solution, optionally sealed within a polymeric film. A gripping portion 58 of the indicator is provided as well to allow the consumer to manipulate the indicator. In the dual indicator embodiment described above, one such

5 formulation for the test portions is as follows:

Chemical Name	Glucose Indicator % (W/W)	Lactose Indicator % (W/W)
10 Potassium Iodide	1.0	1.0
FD&C Blue No. 1 (0.1% aq)	10.0	10.0
15 Water	36.4	16.4
Citric Acid, anhyd	0.6	0.6
20 Sodium Citrate	5.0	5.0
25 Methyl vinyl ether solution (thickener, stabilizer) (10% aq)	10.0	10.0
Polyvinylpyrrolidone (10% aq)	5.0	5.0
30 Horseradish peroxidase (5% aq)	2.0	2.0
Glucose oxidase (1000 units/ml)	30.0	30.0
35 Lactase (3000 LAU/ml)	-	20.0

In use, the consumer will contact an indicator strip with a beverage to be tested. The
40 contacting should be such that each of the glucose and the lactose test squares are each fully
contacted with the sample material. If the lactose test square undergoes a color change that is
different than that of the glucose test square, the presence of lactose in the sample is indicated.

One embodiment of a package for the subject indicator strips is shown in FIG. 3. In the

Figure, the package 80 is of the matchbox type having a cover 82, a plurality of indicators 84, each of which is removeably mounted to a retainer 86. The cover is secured to the retainer using a staple 88, an adhesive or the like. Each of the indicator strips 84 may be torn from the retainer and used to indicate the presence of the desired compounds in a food or beverage sample. As depicted in the Figure, the retainer 86 may include indicators for lactose 90 and caffeine 92 in combination, or the package may include simply a single type of indicator. Additionally, more than a single retainer/indicator combination can be used in each package, thereby allowing multiple indicators with each retainer/indicator combination including a single type of indicator.

In each of the embodiments shown in the figures above, an indicator strip is held by the consumer and contacted with a sample. However, as noted above, it is preferred that the entire indicator not be contacted with the sample, but rather that the sample be placed only on a small portion of the indicator strip. One method for achieving this is by providing a protective jacket or sleeve around the indicator. In another embodiment, however, the indicator may be inserted into (or provided within) a sampling enclosure or housing. One such embodiment is depicted in FIGS. 4a and 4b. In those figures, the sample housing 100 comprises a "U"-shaped tube having a sample collector 102 positioned at one end of the "U". The bend 104 of the "U" forms a sample chamber in which a liquid sample 106 is caused to contact an indicator device 108. If desired, two indicators, for different materials, may be positioned back-to-back within the housing.

In the embodiment depicted in FIGS. 4a and 4b, the indicator 108 is a caffeine indicator as shown in FIG. 1. However, it should be noted that the specific indicator is not intended to be limited to caffeine indicators, but rather, may be any indicator of the type described herein. The housing may be of a single-use type having a single indicator permanently mounted therein, or in the alternative, the leg of the "U" opposite the leg containing the sample collector 102 may be open or openable to allow the indicator to be replaced, thereby providing the housing with a multi-use capability. The housing may be formed of transparent glass; however, a durable, transparent polymer is preferred, as such materials are less likely to suffer breakage during transport and use.

In use, the sample collector 102 is dipped into a sample to be analyzed. For example, the collector 102 may be dipped into a cup of coffee to obtain a small liquid sample 106 having a volume of, for example, approximately 1 milliliter. The sample 106 remains within the collection portion of the device (FIG. 4a) until the device is inverted (FIG. 4b). Once the device

is inverted, the sample fluid 106 flows down to the bend 104 of the "U" and contacts the appropriate portion of the indicator 108. The device is maintained in the position of FIG. 4b until the sample has had an opportunity to wick up the indicator and provide the consumer with the desired assay. Upon completion of the assay, the device may be either discarded (in the
5 single-use embodiment) or emptied and rinsed with fresh water to allow reuse (in the multi-use embodiment).

Having thus described at least one illustrative embodiment of the invention, various alterations, modifications and improvements will readily occur to those skilled in the art. Such alterations, modifications, and improvements are intended to be within the spirit and scope of the
10 invention. For example, the materials employed, as well as their shapes and dimensions may be modified according to the requirements of the device. Accordingly, the foregoing description is by way of example only and is not intended as limiting. The invention is limited only as defined in the following claims and the equivalents thereto.

What is claimed is:

CLAIMS

1. A kit for determining the presence of compounds in food or beverage samples, the kit comprising:
 - a) a plurality of indicator elements, each element having at least one indicator for
5 determining the presence of a compound in a food or beverage sample and an elongate segment by which each indicator element may be manipulated, and
 - b) a retainer removeably joined to each indicator element, to thereby maintain the plurality of indicator elements and the retainer as a single unit until indicator elements are individually removed therefrom.
- 10 2. A kit as in claim 1 wherein the compounds are selected from the group consisting of caffeine and lactose.
3. A kit as in claim 1 which further includes a cover which may be opened to expose the indicator elements.
4. A kit as in claim 2 wherein each indicator element is constructed and arranged to
15 determine the presence of caffeine in the food or beverage sample.
5. A kit as in claim 2 wherein each indicator element is constructed and arranged to determine the presence of lactose in the food or beverage sample.
6. A caffeine indicator which comprises:
 - a) a segment of a material having the ability to wick fluids, the segment having a proximal
20 end and a distal end;
 - b) a sample-receiving portion positioned at the distal end of the segment;
 - c) caffeine labeled with a material capable of causing a visual change in an indicator, the labeled caffeine being positioned proximal to the sample receiving portion;
 - d) caffeine antibodies positioned on the material proximal to the labeled caffeine; and
 - 25 e) an indicator capable of undergoing a visual change in the presence of the labeling material.
7. A caffeine indicator as in claim 6 wherein the material segment comprises an elongated strip.
8. A caffeine indicator as in claim 6 wherein the labeled caffeine is labeled with a material
30 comprising peroxidase.
9. A caffeine indicator as in claim 8 wherein the indicator comprises o-phenylenediamine.

10. A caffeine indicator as in claim 6 wherein at least a portion of the material segment is joined to a support element.

11. A caffeine indicator as in claim 6 wherein at least one of the labeled caffeine, the caffeine antibodies, and the indicator is enclosed within a substantially fluid impermeable, transparent sheath.

12. A caffeine indicator as in claim 6 positioned within a transparent housing.

13. A caffeine test kit which comprises a plurality of caffeine indicator elements each comprising:

a) a segment of a material having the ability to wick fluids, the segment having a proximal end and a distal end;

b) a sample-receiving portion positioned at the distal end of the segment;

c) caffeine labeled with a material capable of causing a visual change in an indicator, the labeled caffeine being positioned proximal to the sample receiving portion;

d) caffeine antibodies positioned on the material proximal to the labeled caffeine; and

e) an indicator capable of undergoing a visual change in the presence of the labeling material;

wherein each of the elements is removeably joined to a retainer, to thereby maintain the plurality of indicator elements and the retainer as a single unit until indicator elements are individually removed therefrom.

14. A caffeine test kit as in claim 13 which further includes a cover which may be opened to expose the indicator elements.

15. A caffeine test kit as in claim 13 wherein at least one of the elements includes a substantially fluid impermeable transparent sheath enclosing at least one of the labeled caffeine, caffeine antibodies, and the indicator.

16. A caffeine test kit as in claim 13 which further includes at least one transparent housing.

17. A lactose indicator which comprises:

a) a segment of a material having the ability to wick fluids, the segment having a proximal end and a distal end;

b) a first indicator for visually indicating the presence of glucose positioned on the material segment; and

c) a second indicator for visually indicating the presence of glucose and lactose positioned

on the material segment substantially adjacent to the first indicator.

18. A lactose indicator as in claim 17 wherein the material segment comprises an elongated strip.

19. A lactose indicator as in claim 17 wherein the first indicator comprises glucose
5 oxidase.

20. A lactose indicator as in claim 17 wherein the second indicator comprises, in combination, glucose oxidase and lactose.

21. A lactose indicator as in claim 17 wherein at least one of the first and second indicators is enclosed within a substantially fluid impermeable, transparent sheath.

10 22. A lactose indicator as in claim 17 positioned within a transparent housing.

23. A lactose test kit which comprises a plurality of lactose indicator elements each comprising:

a) a segment of a material having the ability to wick fluids, the segment having a proximal end and a distal end;

15 b) a first indicator for visually indicating the presence of glucose positioned on the material segment; and

c) a second indicator for visually indicating the presence of glucose and lactose positioned on the material segment substantially adjacent to the first indicator;

20 wherein each of the elements is removeably joined to a retainer, to thereby maintain the plurality of indicator elements and the retainer as a single unit until indicator elements are individually removed therefrom.

24. A lactose test kit as in claim 23 which further includes a cover which may be opened to expose the indicator elements.

25 25. A lactose test kit as in claim 23 wherein at least one of the elements includes a substantially fluid impermeable, transparent sheathing enclosing at least one of the first and second indicators.

26. A lactose test kit as in claim 23 which further includes at least one transparent housing.

27. A test kit for determining the presence of caffeine and lactose in a sample, the test kit
30 comprising at least one caffeine indicator and at least one lactose indicator, each caffeine indicator comprising:

a) a segment of a material having the ability to wick fluids, the segment having a proximal

end and a distal end;

b) a sample-receiving portion positioned at the distal end of the segment;

c) caffeine labeled with a material capable of causing a visual change in an indicator, the labeled caffeine being positioned proximal to the sample receiving portion;

5 d) caffeine antibodies positioned on the material proximal to the labeled caffeine; and

e) an indicator capable of undergoing a visual change in the presence of the labeling material;

and each lactose indicator comprising:

10 a) a segment of a material having the ability to wick fluids, the segment having a proximal end and a distal end;

b) a first indicator for visually indicating the presence of glucose positioned on the material segment; and

c) a second indicator for visually indicating the presence of glucose and lactose positioned on the material segment substantially adjacent to the first indicator.

15 28. A test kit as in claim 27 wherein each of the caffeine indicators is removeably joined to a retainer, to thereby maintain the plurality of indicators and the retainer as a single unit until indicators are individually removed therefrom.

29. A test kit as in claim 27 wherein each of the lactose indicators is removeably joined to a retainer, to thereby maintain the plurality of indicators and the retainer as a single unit until
20 indicators are individually removed therefrom.

30. A test kit as in claim 27 wherein each of the caffeine indicators and each of the lactose indicators is removeably joined to a retainer, to thereby maintain the plurality of indicators and the retainer as a single unit until indicators are individually removed therefrom.

31. A lactose test kit as in claim 27 which further includes a cover which may be opened
25 to expose the indicator elements.

32. A test kit as in claim 27 wherein at least one caffeine indicator or at least one lactose indicator includes a substantially fluid impermeable, transparent sheath which, in the case of the caffeine indicator, encloses at least one of the labeled caffeine, the caffeine antibodies and the indicator, and, in the case of the lactose indicator, encloses at least one of the first and second
30 indicators.

33. A test kit as in claim 27 which further includes at least one transparent housing.

34. The apparatus of any of claims 12, 16, 22, 26, or 33 wherein the housing comprises a "U"-shaped tube.

35. The apparatus of claim 34 wherein the housing includes a sample collector positioned at an end of the "U"-shaped tube.

5 36. A method for determining the presence of caffeine in a beverage which comprises the steps of:

a) providing a caffeine indicator which comprises:

i) a segment of a material having the ability to wick fluids, the segment having a proximal end and a distal end;

10 ii) a sample-receiving portion positioned at the distal end of the segment;

iii) caffeine labeled with a material capable of causing a visual change in an indicator, the labeled caffeine being positioned proximal to the sample receiving portion;

iv) caffeine antibodies positioned on the material proximal to the labeled caffeine; and

15 v) an indicator capable of undergoing a visual change in the presence of the labeling material;

b) contacting the sample-receiving portion with a sample of the beverage to be tested;

c) allowing the sample to be wicked until it contacts the indicator for a period of time sufficient to cause the indicator to provide a visual indication of the presence of caffeine; and

d) observing the indicator to determine whether caffeine is present in the sample.

20 37. A method as in claim 36 wherein a plurality of caffeine indicators is provided, each removeably joined to a retainer, to thereby maintain the plurality of indicators and the retainer as a single unit until indicators are individually removed therefrom, the method including the additional step of removing an indicator from the retainer prior to contacting the sample-receiving portion of that indicator with the beverage sample.

25 38. A method for determining the presence of lactose in a beverage which comprises the steps of:

a) providing a lactose indicator which comprises:

i) a segment of a material having the ability to wick fluids, the segment having a proximal end and a distal end;

30 ii) a first indicator for visually indicating the presence of glucose positioned on the material segment; and

iii) a second indicator for visually indicating the presence of glucose and lactose

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positioned on the material segment substantially adjacent to the first indicator;

b) contacting each of the first and second indicators with a sample of the beverage to be tested;

5 c) allowing the sample to contact each of the first and second indicators for a period of time sufficient to cause the first indicator to provide a visual indication of the presence of glucose and the second indicator to provide a visual indication of the presence of glucose and lactose; and

d) observing the indicators to determine whether lactose is present in the sample.

39. A method as in claim 38 wherein a plurality of lactose indicators is provided, each
10 removeably joined to a retainer, to thereby maintain the plurality of indicators and the retainer as a single unit until indicators are individually removed therefrom, the method including the additional step of removing an indicator from the retainer prior to contacting that indicator with the beverage sample.

1/2

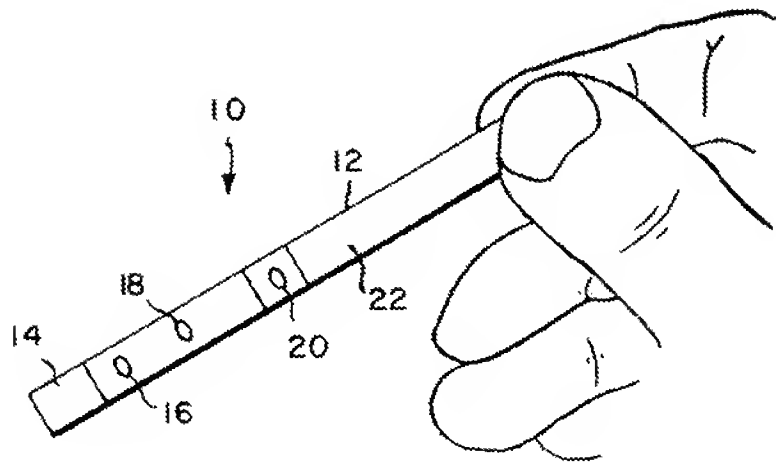


FIG. 1

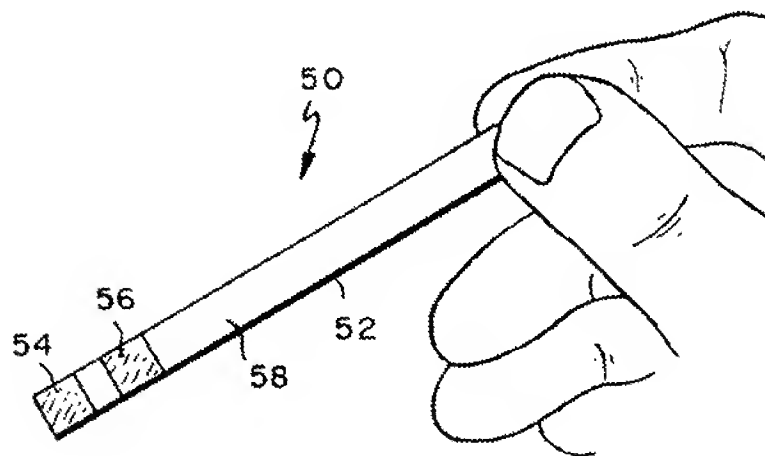


FIG. 2

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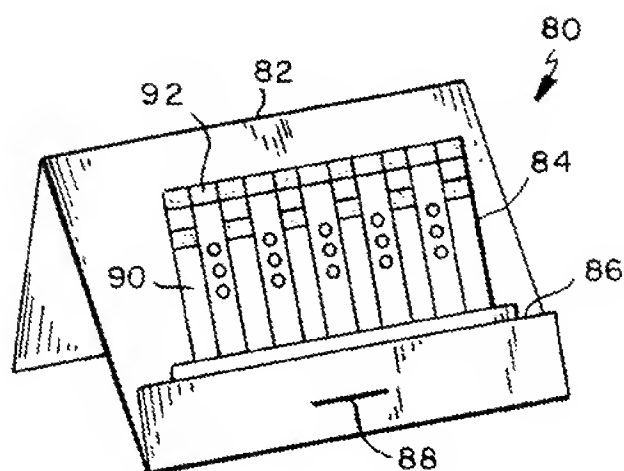


FIG. 3

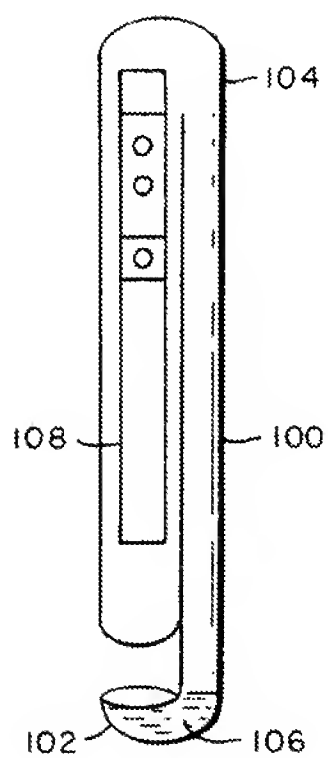


FIG. 4a

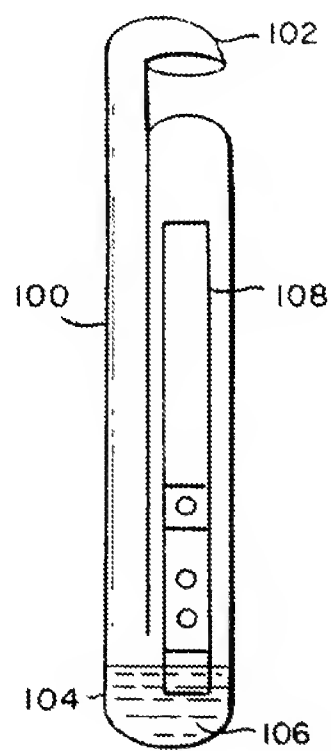


FIG. 4b

INTERNATIONAL SEARCH REPORT

Intern. Application No.
PCT/US 96/02828

A. CLASSIFICATION OF SUBJECT MATTER

G 01 N 31/22, G 01 N 33/02, G 01 N 33/14, C 12 Q 1/25

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

G 01 N, C 12 Q

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US, A. 4 454 094 (BJÖRLING et al.) 12 June 1984 (12.06.84), column 1, line 54 - column 2, line 35; claims.	1, 6, 7, 10-13
A	US, A. 4 647 430 (ZWEIG) 03 March 1987 (03.03.87), column 2, lines 1-55; examples; claims.	1, 6, 7, 10-13, 23, 27
A	GB, A. 2 098 323 (SYBRON CORPORATION) 17 November 1982 (17.11.82), page 1, lines 37-89; claims.	1, 6, 7, 10-13, 23, 27
A	EP, A. 0 158 964 (TERUMO KABUSHIKI KAISHA)	1, 6, 7, 10-13.

☒ Further documents are listed in the continuation of box C.☐ Patent family members are listed in annex.

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Date of the actual completion of the international search
20 June 1996

Date of mailing of the international search report

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INTERNATIONAL SEARCH REPORT

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International Application No.

PCT/US 96/03828

C(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
	23 October 1985 (23.10.85), page 5,6; page 8, paragraph 4 - page 10, paragraph 4; examples; claims. -----	23, 27

ANHANG

zum internationalen Recherchen-
bericht über die internationale
Patentanmeldung Nr.

ANNEX

to the International Search
Report to the International Patent
Application No.

ANNEX E

au rapport de recherche inter-
national relatif à la demande de brevet
international n°

FCT/US 96/02828 SAE 128410

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